Mobility and phytoavailability of Cu, Cr, Zn, and As in a contaminated soil at a wood preservation site after 4 years of aided phytostabilization

Nour Hattab, Mikael Motelica-Heino, Xavier Bourrat, Michel Mench

Mobility and phytoavailability of Cu, Cr, Zn, and As in a contaminated soil at a wood preservation site after four years of aided phytostabilization

Nour HATTAB[a,b*], Mikael MOTELICA-HEINO[a], Xavier BOURRAT[c], Michel MENCH[d]

[a] *ISTO UMR 7327-CNRS/Université d'Orléans Campus Géosciences, 1A, rue de la Férollerie, 45071 Orléans cedex 2, France*

[b] *Laboratoire Sciences du sol, Faculté d'Agronomie, Université de Damas, Damas, Syrie.*

[c] *BRGM, French Geological Survey, 3, Avenue Claude Guillemin - B.P. 36009, 45060 Orléans cedex 02 - France*

[d] *UMR BIOGECO INRA 1202, Ecologie des communautés, Université Bordeaux 1, Bat B8 RDC Est, Avenue des facultés, F-33405 Talence, France*

Keywords: Soil contamination. Copper. Amendment. Phytoavailablity. Phytostabilisation. Trace elements.

Abstract:

The remediation of copper-contaminated soils by aided-phytostabilisation in 16 field plots at a wood preservation site was investigated. The mobility and bioavailability of four potentially toxic trace elements (PTTE), i.e. Cu, Zn, Cr, and As, were investigated in these soils four years after the incorporation of compost (OM, 5% w/w) and dolomite limestone (DL, 0.2% w/w), singly and in combination (OMDL), and the transplantation of mycorrhizal poplar and willows. Topsoil samples were collected in all field plots and potted in the laboratory. Total PTTE concentrations were determined in soil pore water (SPW) collected by Rhizon soil moisture samplers. Soil exposure intensity was assessed by Chelex100-DGT (diffusive gradient in thin films) probes. The PTTE phytoavailability was characterized by growing dwarf beans on potted soils and analyzing their foliar PTTE concentrations. OM and DL, singly and in combination (OMDL), were effective to decrease foliar Cu, Cr, Zn, and As concentrations of beans, the lowest values being numerically for the OM plants. The soil treatments did not reduce the Cu and Zn mineral masses of the bean primary leaves, but those of Cr and As decreased for the OM and DL plants. The Cu concentration in SPW was increased in the OM soil and remained unchanged in the DL and OMDL soils. The available Cu measured by DGT used to assess the soil exposure intensity correlated with the foliar Cu concentration. The Zn concentrations in SPW was reduced in the DL soil. All amendments increased As in the SPW. Based on DGT data, Cu availability was reduced in both OM and OMDL soils, while DL was the most effective to decrease soil Zn availability.

1. Introduction

Phytostabilization is a less invasive, low-cost phytotechnology, singly and in combination with amendments (i.e. aided phytostabilisation) is a potential options to restore the physical, chemical, and biological properties of potentially toxic trace elements (PTTE)-contaminated soils (Bolan et al. 2003; Kumpiene et al. 2008). The fate of PTTE in soils is influenced by physical and chemical reactions between the solid components of soil and the liquid phase (Morel et al. 2006). Soil factors such as pH, soil organic matter (SOM), texture, redox potential and temperature (Alloway 1995) and biological processes controlled by soil microorganisms and plants are key-players in the root zone for the PTTE mobility and bioavailability (Chaignon and Hinsinger 2002; 2003). Roots can indeed modify the PTTE mobility by changing soil pH, electrochemical potentials through element sorption in the apoplast and functioning of membrane transporters and their rhizodeposition or complexation in the rhizosphere, including soluble root exudates and mucilages (Hinsinger 1998, 2001a, b; Lombi et al. 2001; Chaignon and Hinsinger 2002).

Several mineral and organic amendments such as lime, coal fly ashes, phosphates, red muds, compost, biosolids, iron grit and Fe/Mn/Al oxides can improve phytostabilization and production of plant-based feedstock through decrease in the PTTE bioavailability (Lombi et al. 2002; Bolan et al. 2003; Brown et al. 2004; Geebelen et al. 2003; Kumpiene et al. 2008; Mench et al. 2010). Case studies assessing the PTTE mobility and bioavailability in the long-term for contaminated soils managed by (aided) phytostabilization are needed to better define the pros and cons of such management options (Mench et al. 2010). The main thrust of this article was to investigate the effects of the amendments and understand the factors involved in aided phytostabilization of Cu and chromated copper arsenate (CCA) salt contaminated soils by liming and addition of compost. Therefore, this work aimed at assessing the mobility, soil exposure intensity and phytoavailability of Cu, Zn, Cr, and As in top soils (0-25 cm) of 16 field plots at a wood preservation site in Southwest France, four years after their implementation for testing four options of (aided) phytostabilization. These options were either to incorporate compost (OM) and dolomitic limestone (DL), singly and in combination (OMDL), or not into the contaminated soil prior the transplantation of mycorrhizal poplar and willows. The efficiency of the four aided-phytostabilisation options to reduce the mobility and phytoavailability of Cu, Cr, Zn, and As was compared.

2. Material and Methods

2.1. Site, soil sampling and soil characterization

The wood preservation site (6 ha are partially active and 4 ha with historical activities were used for an allotment) is located in the Gironde County (44°43'N; 0°30'O), Southwest France. It has been used for over a century to preserve and store timbers, posts and utility poles and various Cu-based salts were successively utilized (Mench and Bes 2009). Plant communities and soil characteristics were previously assessed. Cu is the main contaminant in topsoils (i.e. 65 to 2600 mg kg^{-1}, Mench and Bes 2009; Bes et al. 2010; 2013). Soil Cu contamination mainly results from washings of the treated timbers. Plant communities in the zone of the field trial included *Agrostis capillaris, Elytrigia repens, Rumex acetosella, Portulaca oleracea, Hypericum*

perforatum, Hypochaeris radicata, Euphorbia chamaescyce, Echium vulgare, Agrostis stolonifera, Lotus corniculatus, Cerastium glomeratum, and *Populus nigra* (Fig. 1a) (Bes et al. 2010). The geological structure of the site consists of two layers. The first one is a mixture of brown sand and gravels, from the medium Pleistocen (Riss); with a depth in the range 2.5- 4.5 m whereas the second layer is composed of marls and decalcification clays related to eroded Stampien materials. Fifteen sub-sites were previously defined (labeled from A to E and P1 to P10) depending on total topsoil Cu concentration (Fig. 1b) (Mench and Bes 2009; Bes et al. 2010). Long-term phytostabilization experiments are established at the P3 and P7 sub-sites. The field trial (150 m²) established in 2006 at the site P1-3, formerly used for stacking treated wood and utility poles (Fig. 1b, Bes 2008; Lagomarsino et al. 2011), has been cultivated as a short rotation coppice including mycorrhizal poplar (*P. nigra* L.) and willows (*Salix caprea* and *S. viminalis)* (Bes 2008). It consists in 16 plots (1 m x 3 m) that have received one of the following four initial treatments, at the beginning of the experiment only, randomly replicated in four blocks: untreated (UNT), dolomitic limestone (DL, 0.2% by air dried soil, w/w, NF U 44 001, 30% CaO and 20% MgO combined with carbonates, 80% < 0.16 cm, Prodical Carmeuse, Orthez, France), compost (OM, 5% w/w), and DL combined with OM (OMDL) (Fig.1c). Thus each treatment was repeated 4 times. Compost derived from composting (9-12 months) poultry manure and pine bark chips (ORISOL, Cestas, France, Bes and Mench 2008). Soil amendments were carefully mixed in the topsoil (0-0.30 m) with a stainless spade. Topsoils (0-25 cm, alluvial origin, Fluviosol) were sampled in April 2010 (average sample made of three sub-samples of 1 kg) with a stainless spade in the 16 plots. Main characteristics of topsoil's at the site P3 are presented in Table 1. Their texture is sandy. Organic matter content is low as well as the cation exchange capacity (CEC). Total soil concentrations were in the common range of French sandy soils for Cr, As, and Zn but total soil Cu was in excess for these coarse sandy soils *i.e.* 35 mg Cu kg^{-1} (Tab. 1, Baize and Tercé. 2002).

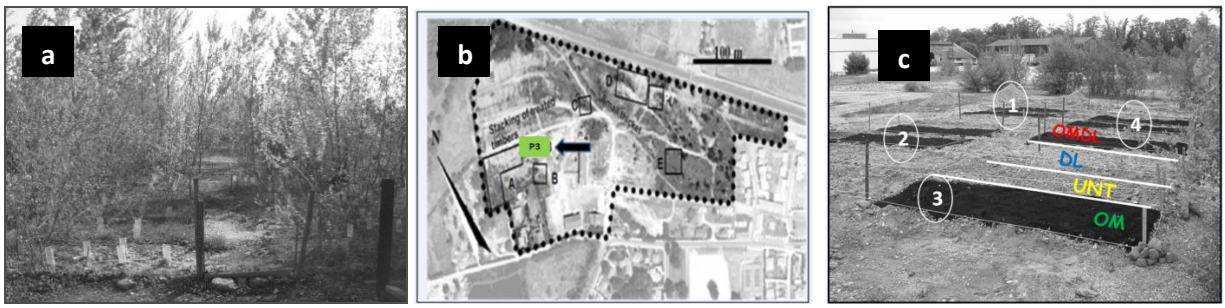

Figure 1. (a) implementation of the plots of the field trial (2006) carried out with soil amendments and mycorrhizal trees since 2006 at the Biogeco phytoremediation platform (adapted from Bes et al.2010), (b) location of the studied site, the arrow indicating the site P3 and (c) Photo of the field plots (P3 sub-site) in April 2010, 4 replicated blocks, each block treated with 4 types of amendements (OM, DL, OMDL and UNT)

Table 1. Main characteristics of the P3 and control soils (0-0.25 m soil layer)

Parameters	Site P3	Control soil	Background values in French sandy soils[a]
Sand %	83.5 ± 1.1	66.5	
Silt %	11.5 ± 0.9	15.5	
Clay %	3.8 ± 1.2	18.0	
C/N	17.2	13.8	
SOM (g. kg^{-1})	15.9	69.9	
CEC (cmol+/kg)	3.49	16.1	
organic C (g. kg^{-1})	9.19	40.4	
total N (g. kg^{-1})	0.534	2.94	
pH	7.0 ± 0.23	7.01	
As (mg.kg^{-1})	9.8	3.6	1.0-25[b]
Co (mg.kg^{-1})	2	2.62	1.4-6.8
Cu (mg.kg^{-1})	674 ± 126	21.5	3.2-4.8
Cr (mg.kg^{-1})	23	17.9	14.1-40.2
Mn (mg.kg^{-1})	181	189	72-376
Ni (mg.kg^{-1})	5	7.46	4.2-14.5
Zn (mg.kg^{-1})	46	50.9	17-48

[a] median and high vibrissae values except for As (Baize. 1997; Baize and Tercé 2002):. [b] common As mean values for all French soil types (Baize and Tercé 2002).

2.2. Germination tests

For each of the 16 plots, 1 kg of soil was potted after sieving (2 mm). Similarly, two samples of an uncontaminated control soil (CTRL) from a kitchen garden (0-0.25 m Gradignan, France) from the same alluvial terrace were taken and potted (Tab 1). Four seeds of dwarf beans (*Phaseolus vulgaris*) were sown in all pots and cultivated for 18 days in controlled conditions (16 h light/8 h darkness regime, 25°C/21°C). The soil moisture was maintained at around 50% of the field water capacity with additions of deionized water after weighing, then the soil moisture was raised to 80% at the beginning of seed germination. At harvest, the dry weight (DW) of bean primary leaves (BLDW) was determined after drying at 70°C. Aliquots of primary leaves (BL) were weighed (35-150 mg) directly into Savillex Polytetrafluoroethylene PTFE 50 mL vessels, 2 mL H_2O and 2 mL of 14 M HNO_3 were added and heated open at 65°C for 2 hours. Then the caps were closed and the containers were left overnight at 65°C (12-14h). Thereafter, they were opened, 0.5 mL of H_2O_2 (30%) was added to each sample and left at 75°C open for 3 hours. Then 1.5 ± 0.5 mL of *fluorhydric* a*cid* (HF, 48%) was added to each sample, caps closed and left at 100°C overnight. Containers were opened and kept at 120°C for 4-5 hours evaporating to dryness, taken off heat, 1 mL HNO_3 + 5 mL H_2O + 0.1 mL H_2O_2 were added to each, gently warmed up (65°C) and after cooling down made up to 50 mL with distilled water**.** Finally, trace element concentrations in digests were determined by ICP-MS (Varian 810-MS) using standard solutions of trace elements diluted from a stock solution1000ppm ±1%/Certified). The accuracy of the metals determination was checked by performing calibrations with a standard reference solution. Strong correlation was found between the measured and the reference results (R^2=0.9992) indicating that the measurement are accurate. After the calibration phase, 4 repeated measurements were performed for each digest. The precision of the trace element

content measurement was assessed by the standard deviation (SD). All foliar element concentrations are expressed in mg kg^{-1} DW. The mineral mass of each PTTE in BL was computed based on their elemental concentrations and the BLDW.

2.3. Characterization of the soil pore water

Soil pore waters (SPW)were extracted from each pot by Rhizon soil moisture samplers (SMS, model MOM, Rhizosphere Research Products, Wageningen, The Netherlands) with a nominal porosity of 0.15 µm after harvesting the dwarf beans. The capped end was inserted into each potted soil during filling. A syringe needle was connected to the female lock and inserted into a 10 mL glass vacuum tube, for extracting the SPW by vacuum (Cattani et al. 2006). Before putting the samplers in the soils, the SMS devices were previously cleaned with 5% HNO_3 and then washed twice with deionised water.

Three SMS devices were placed at 45° in the soils. The soil moisture was maintained at 80% for 15 days. Each device was then let under vacuum for 24 hours for collecting the SPW (~ 30 mL) from all pots (18 soil samples, three SMS/pot). The SPW were stored at 4°C. An aliquot (3 mL) of each SPW was acidified with 0.1 M HNO_3 for measuring the concentrations of Cu, Zn, Cr, and As in the SPW samples by HR-ICP-MS (Element 2, Thermofischer). All the reagents used to prepare the extracting solutions were products of analytical-grade quality (Merck pro-analysis, Darmstadt, Germany). All solutions and dilutions were prepared using doubly deionized water ($18.2M\Omega cm^{-1}$) (Thermo Scientific Barnstead Easy pure II systems). Standard stock solutions of $1000mg.l^{-1}$ of different elements were prepared from metal wires or salts of purity higher than 99.998% (VWR international, BDH Prolabo ICP Standards, Belgium). Diluted standard working solutions were prepared from these on a daily basis. All laboratory glassware and plastic ware were rinsed three times with double deionized water after being soaked in a HNO_3(10%, v/v) bath for 24h. A certified reference material (drinking water) EP-L-3 diluted 1000 times certified by SPS Science (Baie d'Urfé, QC, Canada) was used to assess the precision and accuracy of the analysis of the soil solutions and DGT elution's for Cu, Zn, Cr and As. Measurements lied in the interval of confidence and precision and accuracy was better than 5% RSD.

In all SPW samples, dissolved organic carbon (DOC) was determined by a carbon analyzer (Shimadzu© TOC 5000A), and concentrations of major cations (Na^+, K^+ and Ca^{2+}) and anions (NO_3^-, SO_4^{2-}, and Cl^-) were analyzed by ionic chromatography (Dionex ICS-2000, Sunnyvale, CA using the columns CS16A for measuring cations and AS17 for anions).

2.4. DGT measurements

For measuring available metals in the soils and characterizing the soil exposure, standard cylindrical Chelex 100-DGT (diffusive gradient in thin films) units with an active surface area of 3.14 cm^2 were manually inserted for 24 h directly into the humid topsoil (80%) of each pot. Two DGT probes were inserted per pot. The DGT probes consist of three layers: the first one is a 0.45 µm filter; the second layer is a diffusion layer which consists of a polyacrylamide gel layer and the third one a polyacrylamide gel layer that incorporates a Chelex-100 resin that binds strongly the labile trace metal species (Davison et al. 2000; Ernstberger et al. 2002a, b).

DGT has proved to be an efficient tool to assess the exposure of PTTE in contaminated soils (Zhang et al., 2001).

DGT continuously accumulates metals on the resin gel during deployment. The total mass of each metal (*M*) accumulated per unit area over the deployment time (*T*) is given by integrating the flux over the deployment time (eq. 1):

$$M = \sum_{t=0}^{T} F(T)dt \qquad \text{(eq. 1)}$$

The total mass of each metal *(M)* is determined analytically through the area exposed to the solution *(A)* by measurement of the eluent concentration (*C*e) after elution of the resin gel (volume, *V*gel) with 1 M HNO_3 (volume, *V*HNO3),

$$M = C_e\,(V_{HNO3} + V_{gel})/fe \qquad \text{(eq. 2)}$$

With f=0.8 for Cu, Cr and Zn.

The strong binding of metals in the resin gel leads to the creation of a linear concentration gradient in the diffusive gel. This gradient depends on several factors such as the interfacial concentration of labile trace metal species, C_i and the thickness of the diffusion layer, Δg (cm), All these factors determines the flux, $F(t)$, of metal from the soil to the resin-gel according to Flick's first law (eq. 3):

$$F(t) = \text{Ød}Dd\frac{Ci(t)}{\Delta g} \qquad \text{(eq. 3)}$$

where Ød is the porosity of the diffusion gel and Dd (cm^{-2} s^{-1}) is the diffusion coefficient of the labile trace metal species in the diffusion layer. The diffusion coefficients were taken from Zhang et al. (2001).

The averaged interfacial concentration, C_{DGT}, or available concentration can be calculated from *M* (eq. 4)

$$CDGT = \frac{M\Delta g}{\text{Ød}Dd} \qquad \text{(eq. 4)}$$

For each metal, the division of the available concentration (C_{DGT})on the total concentration measured in the SPW, C_{SPW}, give the ratio, *R*, which indicates the extent of the depletion of soil pore water concentrations at the DGT interface (eq. 5)

$$R = C_{DGT}/C_{SPW} \qquad \text{(eq. 5)}$$

For each metal, the mass accumulated in the resin-gel layer was determined after extraction of the resin gel by 1 mL of HNO_3 5% for 24h. This solution was further diluted 10 times and analyzed by HR-ICP-MS (Element 2, Thermo Fischer) for determining metal concentrations (Cu, Cr and Zn).

2.5. Statistical analysis

Analysis of variance (ANOVA), Tukey Post Hoc test (Statistica) and Pearson correlation coefficients (linear regression) (significance level, $p<0.05$) were performed on the total SPW concentrations, DGT concentrations, R ratios, foliar element concentrations, foliar mineral masses of elements and leaf DW yields to evaluate the treatment influence. All analytical determinations were performed in four replicates. Differences were considered statistically significant at $p < 0.05$. All statistical analyses were performed using the statistical software Statistica (version 6).

3. Results and discussion

3.1. Soils and soil pore waters

Table 2 shows the physico-chemical parameters of soils (i.e. pH, EC and TOC) and of the SPW (i.e. DOC and major cation and anion concentrations) depending on soil treatments, i.e. phytostabilization (UNT) and aided phytostabilization (OM, DL, and OMDL). Aided phytostabilization based on treatment incorporation into the soil followed by tree transplantation increased slightly the soil pH from 7.16 up to 7.45 in the treated soils compared with the untreated one, mean soil pH values following the decreasing order: DL > OMDL > OM > UNT > CTRL. However, differences in the soil pH and TOC (0.53-1.68%) and in DOC of SPW (29-63 mg C. L^{-1}) were only significant between the contaminated soils (UNT, OM, DL, and OMDL) and the uncontaminated control soil (CTRL) ($P=0.0019$, $P= 1.45E^{-6}$, $P= 0.0013$). They were insignificant between the treated and untreated contaminated soils. The soil EC values varied in the 157-192 µS cm^{-1} range but did not differ between all investigated soils.

Four years after, amendments added to the contaminated soils had little influence on the cation concentrations in the SPW (Tab. 2). The SPW Mg^{2+} concentrations varied from 10.9 mg L^{-1} (Unt) to 41.3 mg L^{-1} (DL) and was significantly higher for the DL soil compared to the UNT soil ($P=0.014$), The SPW Na^{+}, K^{+}, and Ca^{2+} concentrations were respectively in the 8-38 mg L^{-1} Na^{+}, 16-49 mg L^{-1} K^{+}, and 87-272 mg L^{-1} Ca^{2+} ranges, but differences between soils were insignificant. For the anion concentrations in the SPW, Cl^{-}, NO_3^{-}, and SO_4^{2-} varied respectively between 9.6-42.9 mg L^{-1}, 300-914 mg L^{-1}, and 13.9-42.7 mg L^{-1}, without significant differences across the soil series (Tab. 2).

Metal concentrations in SPW generally mirror root exposure to metals (Sauvé 2003; Tandy et al. 2006; Forsberg et al. 2009). Soil pore waters collected from the potted contaminated and uncontaminated soils showed significant differences in total metal concentrations (Fig. 2). On this soil series, total PTTE concentrations in the SPW were generally in decreasing order: Cu > Zn > As > Cr (mean values in µg L^{-1} on the soil series: Cu = 676, Zn = 22, As = 3.0, and Cr = 0.68). This ranking reflected total PTTE concentrations in the soils, except that total soil Cr was higher than total soil As (Tab.1).

Table 2. Physico-chemical characteristics of the soils and soil pore water. Values are mean ± standard deviation (n=4). Different letters indicate a significant difference ($p<0.05$)

Soils	pH	EC (µS. cm^{-1})	TOC %	DOC (mg C.L^{-1})	Cations (mg. L^{-1})				Anions (mg. L^{-1})		
					Na^+	Mg^{2+}	K^+	Ca^{2+}	Cl^-	NO_3^-	SO_4^{2-}
UNT	7.16±0.12 a**	161±64.0 NS	0.64±0.02 a***	29.4±8.80 a**	21.0±6.60 NS	10.9±6.3 a*	21.9±19.6 NS	156±56.5 NS	16.9±2.10 NS	495±207 NS	19.6±4.2 NS
OM	7.27±0.38 a**	168±51.0 NS	1.68±0.33 a***	38.0±7.10 a**	38.7±35.3 NS	22.9±14.0 ab*	41.4±26.6 NS	167±114 NS	42.9±45.7 NS	548±35 NS	39.3±28.4 NS
DL	7.45±0.15 a**	157±16.0 NS	0.53±0.05 a***	39.4±6.50 a**	28.2±11.9 NS	41.3±18.9 b*	24.7±18.4 NS	273±148NS	26.7±17.8 NS	914±527 NS	42.7±28.0 NS
OMDL	7.32±0.11 a**	192±39 .0 NS	0.72±0.09 a***	40.9±4.70 a**	19.5±5.70 NS	21.8±9.40 ab*	16.4±3.8 NS	87.5±41.7 NS	18.3±7.20 NS	301±166 NS	18.4±7.7 NS
CTRL	6.45±0.07 b**	169±22.0 NS	0.62±0.30 b***	63.06±1.70 b**	8.6±8.50 NS	23.2±10.7 ab*	49.7±7.9 NS	113.7±60.8 NS	9.6±12.8 NS	*ND*	13.9±9.8 NS

0.01<(*)P <0.05, 0.01>(**) p >0.001, (***) p< 0.0001, (NS) insignificant difference; ND: not determined. Unt : untreated, OM : Compost, DL : Dolomitic limestone, OMDL : compost and dolomitic limestone, CTRL : uncontaminated control soil. EC: electrical conductivity, TOC: total organic carbon; DOC: dissolved organic carbon

3.1.1. Soil Cu exposure

The addition of OM and OMDL numerically increased 2 fold and 1.3 fold respectively the total dissolved Cu concentration in the SPW (Cu_{spw}) whereas the DL treatment slightly decreased it (0.8 fold) compared to the UNT soil; however only the OM soil differed from the UNT soil (Fig. 2a). The Cu_{spw} value of the CRTL soil was 58%, 79%, 52% and 67% lower than those of the UNT, OM, DL and OMDL soils, respectively, i.e. 2-5 fold less than for these contaminated soils. Compost (OM) incorporation into the contaminated soil significantly increased Cu_{spw} compared to other amendments ($P=9.0\ E^{-19}$) and the uncontaminated soil (CRTL) ($P=1.12\ E^{-10}$) (Fig. 2a). All amendments significantly decreased the available Cu concentration (Cu_{DGT}) in the contaminated soils by roughly a factor 2 ($P= 1.38\ E^{-14}$) (Fig. 2e). Cu_{DGT} peaked in the UNT soil and was the lowest in the CTRL soil ($P = 5.4\ E^{-16}$). In contrast with Cu_{spw} (Fig. 2a), Cu_{DGT} was lower in the OM and OMDL soils than in the DL soil (Fig. 2e). The Cu_{spw} values in the OM and OMDL soils were respectively 7 fold and 5 fold higher than the Cu_{DGT} values, i.e. (in µg Cu L^{-1}) OM: 1065 and 157 respectively; OMDL: 665 and 147, respectively) (Fig.2. a and e). Similar comparison showed a 2-fold factor for the UNT soil (519 µg Cu L^{-1} and 280 µg Cu L^{-1}) and the DL soil (456 µg Cu L^{-1} and 197 µg Cu L^{1}).

On the whole soil series, the Cu_{spw} values were not correlated to the Cu_{DGT} ones (r = -0.16 the correlation in the contaminated and the control soil, r = -0.57 the correlation calculated in the contaminated soil without the control one), but highly positively correlated with the soil TOC values (r = 0.86 "contaminated soil with control one") (Tab. 3). The correlation between Cu_{spw} and TOC was even stronger when only the Cu-contaminated soils are considered (r = 0.93) (Tab. 4). The Cu_{spw} did not significantly correlate to soil pH (r = 0.35 "in the presence of control soil, r=-11 only in contaminated soils, Tab. 3), The correlation was significant and negative between Cu_{DGT} and the DOC measured in the SPW (r = -0.68) (Tab. 3,4).

Table 5 presents the R ratio ($R_{Cu} = Cu_{DGT}/Cu_{spw}$) values for the whole soil series. The R_{Cu} values of all amended soils were lower than that for the UNT soil. The soil amendments reduced R_{Cu} and led to the increasing order: OM < OMDL < DL < UNT, this reduction reaching respectively 73% (OM), 59% (OMDL), and 20% (DL) compared to the UNT soil. The R_{Cu} of the control soil was 35% lower than that of the UNT soil.

The Cu phytotoxicity in soils depends mainly on its solubility and chemical speciation, which are influenced by its sorption onto mineral and soil organic matter (SOM) (Garrido et al. 2005). Here the total dissolved Cu concentration in SPW increased significantly in the OM-amended soils and slightly in the OMDL soils (Fig. 2a). The DOM (dissolved organic matter) may mobilize soil Cu and bound it in the SPW (Beesley and Dickinson 2011). However, four years after adding OM and OMDL, DOC in SPW as well as soil pH were rather similar in both compost-amended soils and the DL soil (Tab. 2). One week after compost incorporation into the soil, Cu mobility can be reduced by 71% (Ruttens et al. 2006). The SOM and DOM in the SPW react with Cu, and their complexes modify Cu solubility, chemical species, and re-supply from soil bearing phases (Ashworth and Alloway 2007). Fulvic acid bound Cu in the SPW increases its mobility from the solid phase to the liquid phase of soils, notably in compost-amended soils (Hsu and Lo 2000). Soluble Cu may also increase in the SPW in the presence of DOM whereas high molecular mass organic compounds can sorb Cu (McBride and Martinez, 2000; Docekal et al. 2005). After one year, Cu bound to organic matter increased 3-fold in compost-amended soils at this wood preservation site (Lagomarsino et al. 2011). The Cu–SOM complexes, particularly with non-soluble, high molecular mass organic acids can decrease Cu phytoavailability (Chirenje and Ma 1999; Balasoiu et al. 2001; Bolan and Duraisamy 2003). Regulation of Cu mobility from soil to plants by SOM in relation to the ligand functional groups was suggested by Hsu and Lo (2000) and Thakali et al. (2006). In calcareous soils OM may reduce the retention of Cu by $CaCO_3$ and increase its solubility in the soil (Saha et al.1991) as in the OMDL soil (Fig. 2a). The insignificant, small reduction in Cu_{spw} (0.8 fold) in the DL soil can be explained by the slight increase in soil pH (Sauve´ et al. 1997). The DL addition may limit Cu_{spw} by Cu precipitation and complexation and increasing the Cu sorption on solid bearing phases such as organic matter, clays, Fe/Mn (hydr) oxides, carbonates and phosphates (Filius et al. 1998; Kabata-Pendias and Pendias 2000; Garrido et al. 2005 Kumpiene et al. 2006; Lagomarsino et al. 2011). The DL addition induced a similar trend when OM and OMDL soils were compared (Fig. 2a). Similarly, liming soils from pH 4.6 to 6.9 can decrease the water soluble and exchangeable Cu fraction from 11.7 to 4.6 mg kg^{-1} in sludged soils (Brallier et al. 1996).

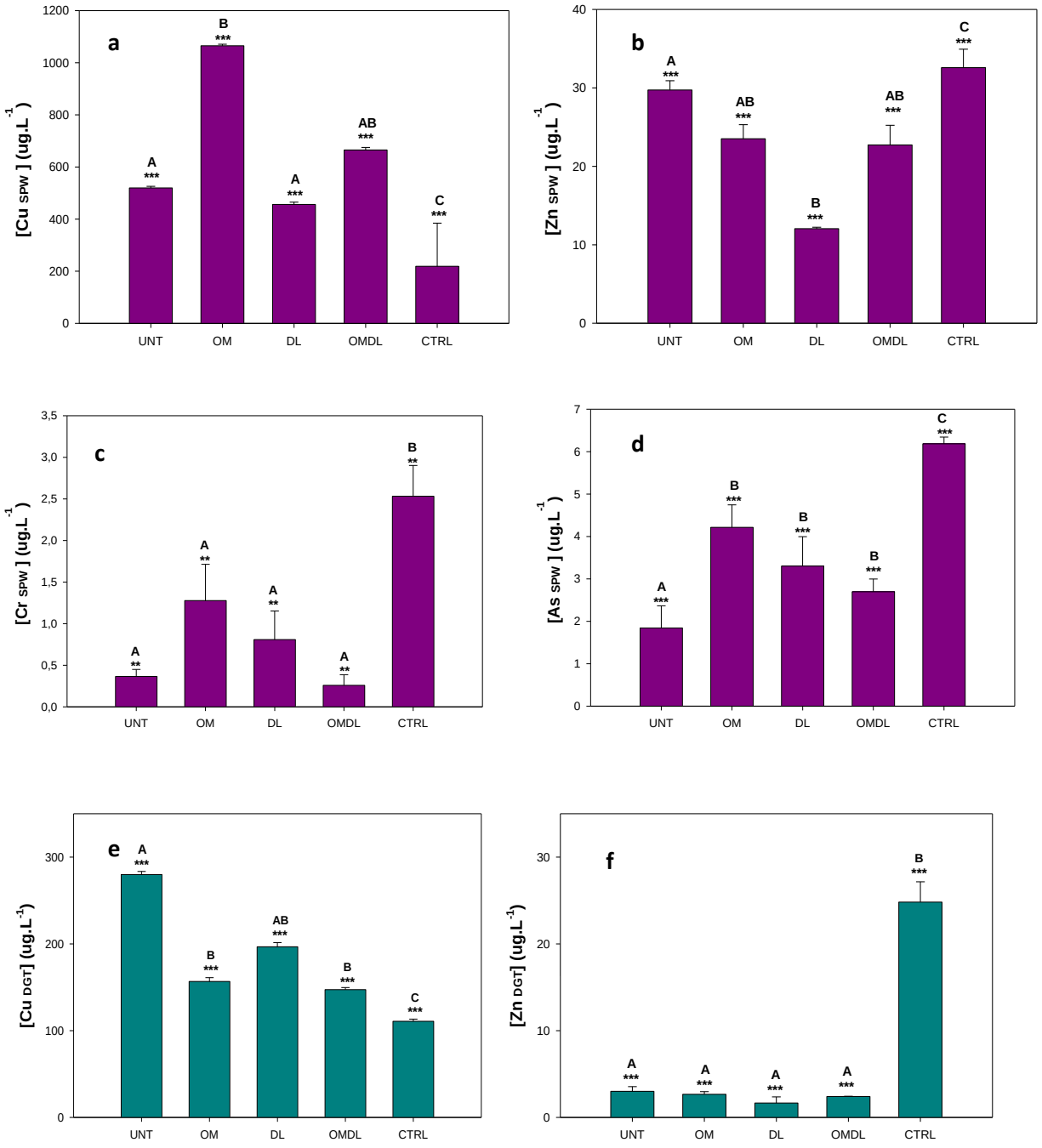

Figure 2. Concentrations of Cu, Zn, Cr, and As in the soil pore waters and intensity of Cu and Zn exposure in the soils determined by DGT. Values are mean ± standard deviation (n=4). Different letters on bar graphs indicate a significant difference ($p<0.05$)

Table 3: Pearson's correlation coefficients between the parameters of soils and soil pore waters of the contaminated and control soil.

	Amendement	pH	EC (µS. cm^{-1})	TOC (%)	DOC (mg.L^{-1})	[Cu $_{tot}$]	[Cu $_{spw}$]	[Cu $_{DGT}$]	R
Amendement	1.00								
pH	-0.56**	1.00							
EC (µS. cm^{-1})	0.10 ^{NS}	-0.17 ^{NS}	1.00						
TOC (%)	-0.26 ^{NS}	0.10 ^{NS}	0.10 ^{NS}	1.00					
DOC (mg.L^{-1})	0.83***	-0.47**	0.10 ^{NS}	-0.03 ^{NS}	1.00				
[Cu $_{tot}$]	-0.65**	**0.61****	0.24 ^{NS}	0.14 ^{NS}	**-0.65****	1.00			
[Cu $_{spw}$]	-0.49**	0.35 ^{NS}	0.09 ^{NS}	**0.86*****	-0.33 ^{NS}	0.46**	1.00		
[Cu $_{DGT}$]	-0.75***	0.23 ^{NS}	-0.18 ^{NS}	-0.29 ^{NS}	**-0.68****	0.30 ^{NS}	-0.16 ^{NS}	1.00	
R	-0.23 ^{NS}	-0.06 ^{NS}	-0.21 ^{NS}	**-0.69****	-0.26 ^{NS}	-0.12 ^{NS}	-0.71***	0.80***	1.00

Significance level: 0.01<(*)P <0.05, 0.01>(**) p >0.001, (***) p< 0.0001, (NS) insignificant difference.

TOC: total organic carbon, Cu_{DGT}: interfacial Cu concentration determined by DGT, Cu_{spw}: total Cu concentration in the soil pore water; DOC: dissolved organic carbon, EC: electrical conductivity, Cu $_{tot}$: total soil Cu, Amendement: the four types of soil treatments (UNT, OM, DL, and OMDL).

Table 4: Pearson's correlation coefficients between soil and soil pore water parameters in the contaminated soil (without control).

	Amendement	pH	EC (µS. cm^{-1})	TOC (%)	DOC (mg.L^{-1})	[Cu $_{tot}$]	[Cu $_{spw}$]	[Cu $_{DGT}$]	R
Amendement	1.00								
pH	0.35 ^{NS}	1.00							
EC (µS. cm^{-1})	0.21 ^{NS}	-0.27 ^{NS}	1.00						
TOC (%)	-0.21 ^{NS}	-0.07 ^{NS}	0.11 ^{NS}	1.00					
DOC (mg.L^{-1})	0.54*	0.31 ^{NS}	0.18 ^{NS}	0.16 ^{NS}	1.00				
[Cu $_{tot}$]	0.11 ^{NS}	0.62*	-0.08 ^{NS}	0.24 ^{NS}	0.46 ^{NS}	1.00			
[Cu $_{spw}$]	-0.08 ^{NS}	-0.11 ^{NS}	0.09 ^{NS}	**0.93*** **	0.14 ^{NS}	0.23 ^{NS}	1.00		
[Cu $_{DGT}$]	-0.76**	-0.25 ^{NS}	-0.21 ^{NS}	-0.43 ^{NS}	-0.55*	-0.32 ^{NS}	**-0.57***	1.00	
R	-0.47 ^{NS}	-0.09 ^{NS}	-0.21 ^{NS}	-0.70**	-0.41 ^{NS}	-0.26 ^{NS}	-0.84***	0.91***	1.00

Significance level: 0.01<(*)P <0.05, 0.01>(**) p >0.001, (***) p< 0.0001, (NS) insignificant difference.

TOC: total organic carbon, Cu $_{DGT}$: interfacial Cu concentration determined by DGT, Cu $_{spw}$: total Cu concentration in the soil pore water; DOC: dissolved organic carbon, EC: electrical conductivity, Cu $_{tot}$: total soil Cu, Amendement: the four types of soil treatments (UNT, OM, DL, and OMDL).

Table 5: Capacity of the solid phase to resupply Cu, Zn, and Cr to the pore water: Ratio of the available concentration determined by DGT vs. total dissolved soil pore water concentration (C_{DGT}/ C_{spw})

Amendment	R(Cu)	R(Zn)
UNT	0.54 **a**±0.01	0.10 **a** ±0.02
OM	0.15 **b**±0.00	0.12 **a** ±0.01
DL	0.43 **ab**±0.02	0.16 **a** ±0.60
OMDL	0.22 **c**±0.01	0.15 **a**±0.03
CTRL	0.34 **abc**±0.01	0.76 **b** ±0.02

R > 0.95: high capacity of the solid phase to resupply the pore water ; R < 1 : the DGT device is supplied only by the diffusion of solutes through the pore water which becomes progressively depleted. Other R values : partial resupply from the solid phase, but insufficient to sustain fully pore water concentrations. Mean values in a column followed by the same letter did not differ at the 5% level

3.1.2. Soil Zn exposure

The Zn concentration in the SPW (Zn_{spw}) was reduced for the amended soils, i.e. OM: 21%, OMDL: 24% and DL: 59% ($P= 5.35E^{-05}$) compared with the UNT soil (Fig. 2b). The DL influence was the most significant. The Zn_{spw} value for the CTRL soil was higher than for all the contaminated soils, i.e. UNT: 1.09 fold, DL: 2.7 fold, and OM and OMDL: 1.4 fold ($P= 5.94E^{-07}$). In soils amended with 5% compost, with and without 5% cyclonic ashes, the Zn mobility was reduced after one week by 87% and 96% respectively compared with the UNT soil (Ruttens et al. 2006). Limestone, biosolids, cyclonic ashes, iron grit and red muds can also decrease soil Zn mobility (Brown et al. 2005). For instance the decrease in Zn_{spw} in compost- and biochar-amended soils over a 60-day field exposure was due to the Zn presence mainly in water-soluble fractions whereas other elements such as Cu and As formed more stable complexes in the soil (Beesley et al. 2010). Conversely, Zn was immobilized in an acid soil by humic acids isolated from organic materials, whilst Cu mobility was enhanced by the same humic acids (Clemente and Bernal 2006). Zinc is generally relatively insoluble at pH>7 (Ross 1994), so the higher Zn_{spw} value in the CTRL soil would be related to its lower soil pH (Tab. 1). Soluble Zn concentrations in SPW from environmentally exposed green waste compost are generally low compared to other PTTE such as Cu and As (Beesley and Dickinson 2010). The OM addition rate may influence Zn mobility and increase negatively-charged adsorption sites in the OM-treated soils (Hartley et al. 2010). In González et al. (2012), an OM addition rate of 6%, compared to 5% in our experiment, decreased Zn_{spw} while it increased at 2%.

Red mud was more effective than limestone and furnace slags to decrease shoot Zn concentration of lettuce (Lee et al. 2009). Reduced concentrations of soluble and extractable Zn in the amended soils were partly attributed to increase in the soil pH. Addition of alkaline materials such as coal fly ash and red mud also decreased Zn leaching by 99.7% and 99.6%, respectively (Ciccu et al. 2003), compared to 59% in our study. Large reductions in Zn extractability (up to 21.9%) and phytoavailability can be obtained from alkaline organic treatments, i.e. lime-stabilized biosolid and N-Viro Soil, by forming metal-carbonate precipitates (Basta et al. 2001).

The DL treatment decreased Zn_{DGT} by 45% compared to the UNT soil, being more effective than OM (11%) and OMDL (20%) (Fig. 2f). Zn_{DGT} was 8- 12 times higher in the uncontaminated soil than in the contaminated ones ($P=2.30\ E^{-08}$) (Fig. 2f).

Zn_{spw} in the OM, DL and OMDL treated soils was respectively 9, 7 and 9.5 fold higher than Zn_{DGT}. This ratio peaked up to 10 for the UNT soil (29.7 and 3, µg Zn L^{-1}, respectively). The DOC correlated with Zn_{DGT} (r=0.776) in the contaminated and control soils, and Zn_{DGT} with Zn_{spw} as well (r=0.57). The Zn_{DGT}:Zn_{SPW} ratio (R) was roughly 7 fold higher in the control soil than the UNT soil, the solid phase of this last one having a low capacity to resupply Zn the pore water (Tab. 5). The R value was numerically slightly lower in the UNT soil than in the treated soils and peaked in the DL soil, but differences were not significant.

In another study the Zn availability in a contaminated soil can be reduced after the incorporation of various amendments (i.e. limestone, biosolids, cyclonic ashes, iron grit and red muds), but ecosystem services

were not fully restored due the residual Zn bioavailability (Brown et al. 2005). Increase in the soil pH, due to organic and inorganic amendments, was suggested in line with Zn precipitation and sorption on mineral phases (Lee et al. 2009). Similar findings were reported with six cost-effective amendments ($CaCO_3$, iron grit, fly ash, manure, bentonite and bone meal) for Cd, Zn and Pb leaching and phytoavailability (Houben et al. 2012). Here, the DL incorporation into the soil slightly increased soil pH (Tab. 2), significantly decreased Zn_{SPW}, but did not change Zn_{DGT} which was initially low.

3.1.3. Soil Cr exposure

The total dissolved Cr concentration in the SPW of contaminated soils (Cr_{spw}) (in µg. L^{-1}) varied between 0.3 (OMDL) and 1.3 (OM). However this variation was not significant for the treated (OM, DL and OMDL) and untreated (UNT) contaminated soils. In contrast the control soil (CTRL) presented the highest total dissolved Cr concentration in the SPW (Cr_{spw}) (in µg. L^{-1}) (2.5), this concentration was significant compared with the others contaminated soils (p=0.002)(Fig. 2c).

Cr_{spw} concentration was related to TOC of the treated and untreated contaminated soils (r=0.65). Similarly the Cr_{spw} concentration was related to the DOC (r= 0.82) measured in the contaminated (UNT, OM, DL and OMDL) and uncontaminated soils (CTRL). Cr_{DGT} was below the detection limit for all contaminated soils. Thus the $Cr_{DGT:}$:Cr_{SPW} ratio was Zero in the four type of soil.

Cr mobility depends on several key-factors such as soil pH, clay mineral content, competing major ions, and complexing agents (Pantsar-Kallio et al. 2001). The oxidation state of Cr in contaminated soils is an important indicator of toxicity and potential mobility. Chromium in the hexavalent Cr (VI) state is highly toxic and soluble, whereas the trivalent state Cr (III) is much less toxic and relatively insoluble. Increased Cr_{spw} after a single addition of both organic or inorganic amendment, and correlation between the soluble Cr fraction and the total and dissolved organic matter in the soil were previously reported (Kumpiene et al. 2008; Hartley et al. 2010). As Cr (III) is relatively insoluble and resistant to leaching (Palmer and Puls 1984), the fraction we measured may include chromate (hexavalent chromium; Cr (VI)) which is more mobile under alkaline to slightly acidic conditions (e.g. pH=7.27±0.38 in the OM soil) Kimbrough et al. 1999). Higher chromate mobility may be due to the presence of other competing anions which were abundant in the compost (Jardine et al. 1999). Based on Pantsar-Kallio et al. (2001) several alkaline materials, e.g. fly ash, hydroxyapatite, and $CaCO_3$, can increase soil pH above neutral, which favor the oxidation of Cr(III) to Cr(VI), and thus enhances the Cr mobility and uptake by roots (Rai et al. 2004). This agrees with the higher Cr_{spw} value measured in the DL soil. Barnhart et al. (1997) indicated lower sorption of Cr species at higher pH. Decline in Cr_{SPW} in the OMDL soil can be due to the capacity of soil organic matter (SOM) to reduce Cr (VI) to Cr (III), which is more stable in the soil, and the negative functional groups on SOM surface which can increase Cr (III) adsorption (Bolan et al. 2003; Banks et al. 2006; Jiang et al. 2008). Simultaneously, the DL addition into the OMDL soil slightly increased soil pH (Tab. 2), which may promote Cr (III) sorption by SOM.

411 Available Cr fraction determined by DGT was not detected in the contaminated topsoil's, suggesting that
412 soil Cr may be mainly under the Cr (III) form, which is more stable or not available to the DGT probe
413 (Ernstberger et al. 2002). Even though chromates are not expected to bind to the chelex resin due to their anionic
414 form, they can diffuse in the gel layer but this one was not analyzed.
415

416 ### 3.1.4. Soil As exposure

417 The total dissolved As concentration in the SPW ($As_{spw,}$ in µg. L^{-1}) increased from 1.8 to 4.2 in all
418 amended soils compared to the UNT soil, and ranked as: OM > DL > OMDL > UNT (Fig. 2d). The As_{spw}
419 peaked in both the OM soil (P=0.0003) and the CTRL soil, which had a relatively high SOM content. As_{spw} well
420 correlated with TOC for our soil series (r= 0.69) and DOC concentrations in soil pore water (r= 0.73).

421 All amendments incorporated into the soil increased the As mobility in the treated soils but As_{SPW}
422 remained below the control soil value (Fig. 2d). Influences of organic and inorganic amendments on the
423 mobility, availability and phytotoxicity of soil As in the soil are controversial. Increases in As mobility after
424 dressing of organic matter are reported (Mench et al. 2003). DOM generally presents in SPW in anionic form
425 and compost-borne anions such as phosphates may compete with As for sorption sites such as Fe oxides (Sadiq
426 1997)). This could enhance As leaching from the soil material (Lombi et al. 2000). In the OMDL treatment,
427 changes in the soil pH (Tab. 2) and addition of Ca and Mg, may promote precipitation of Ca- and Mg
428 phosphates and arsenates, metal arsenates such as those of Cu(II), which are less soluble and more stable in the
429 neutral pH region than Ca arsenates, and may limit DOM influence. In CCA-spiked mineral soils, 92% of total
430 As is As(V), which is less mobile and less toxic, while the proportion of mobile, toxic and bioavailable As(III)
431 in CCA-spiked organic soils increased to one third of the total soil As (Balasoiu et al. 2001). Other mechanisms
432 have been suggested: (1) organic anions may block As adsorption sites (Carey et al. 1996) and (2) formation of
433 soluble As-organic complexes (Chen et al. 2006; Dobran and Zagury 2006).

434 DL incorporation into the contaminated soil increased the mobility of As by 2 fold (Fig. 2d), confirming
435 previous findings (Seaman et al. 2003; Mench et al. 2003). Alkaline materials such as lime, dolomitic limestone,
436 fly ashes, and hydroxyapatites, are indeed undesirable in As-contaminated soils as they can increase As release
437 from the soil to the SPW, due to the higher As mobility at a higher pH range. However, lime could slightly
438 reduce As leaching in soil through possible formation of As–Ca complexes. Calcium hydrogen arsenate
439 ($CaHAsO_4$) and calcium arsenate ($Ca_3(AsO_4)_2$) can precipitate in the Ca presence under oxidizing and moderate
440 pH conditions (Porter et al. 2004).
441

442 ## 3.2. Biomass of primary leaves and PTTE phytoavailability

443 Liming and organic amendments, singly and in combination, can increase plant yields and reduce the
444 plant exposure to Cu and Zn in metal-contaminated soils (Brallier et al. 1996; Sanchez-Monedero et al. 2004; Su
445 and Wong 2004; Bes and Mench 2008). In our study the BLDW of Cu and Zn was numerically higher for all

446 contaminated soils compared to the CTRL soil (Tab. 6). For the contaminated soils the lowest BLDW value was
447 for the UNT beans: based on the UNT plants, the OM, OMDL, and DL treatments numerically enhanced BLDW
448 by 50%, 44%, and 23% respectively (Tab. 6).

449 The UNT beans had higher foliar Cu, Cr, As, and Zn concentrations than the other beans (Tab. 6). Foliar
450 metal concentrations were reduced in the OM, OMDL and DL plants compared to UNT plants, which agreed
451 with Bes and Mench (2008). The addition of dolomite residue and to a lesser extent gypsum and
452 phosphogypsum can also reduce both soil Cu exposure and plant Cu concentration (Garrido et al. 2005). Foliar
453 Cu, Cr, and As concentrations of plants grow on the contaminated soils exceeded those of CTRL plants, except
454 foliar Zn concentration which was similar. Differences between the treated soils and the UNT soils and between
455 the contaminated soils and the CTRL soil were significant for Cu, Cr, As and Zn, except for foliar Zn
456 concentration between contaminated and CTRL soils. Except for CTRL beans, foliar Cu concentration exceeded
457 its upper critical threshold value (in mg kg^{-1} DW: 15-30 (MacNicol and Beckett 1985); 20 (Kabata-Pendias and
458 Pendias 2000)). Foliar Cu concentration was predicted by Cu_{DGT} (r=0.78) and was correlated to soil
459 pH (r= 0.62). Upper critical threshold values for foliar Cr, As and Zn concentrations, i.e. 1-5, 1-5, and 100-450
460 mg kg^{-1} DW, respectively (MacNicol and Beckett 1985; Kabata-Pendias and Pendias 2000) were just reached in
461 the UNT plants and in overall decrease in bean growth mainly mirrored Cu phytotoxicity confirming previous
462 findings (Bes and Mench 2008).

463 Total element amount in primary leaves (µg $plant^{-1}$), so-called mineral mass, was computed for Cu, Cr, Zn
464 and As by multiplying the foliar element concentration (µg kg^{-1} DW) by the leaf biomass (µg DW $plant^{-1}$). For
465 Cu, Cr and As, mineral masses of UNT plants were higher than those of the CTRL plants (Fig. 3). Soil treatment
466 influenced mineral masses for Cr, Cu and As but not for Zn. This reflected a dilution effect of the biomass on
467 Zn concentrations, i.e. foliar Zn concentration = $176.93e^{-10,61}$ foliar DW yield (R^2 = 0.93). Compared to the UNT
468 plants, mineral masses of OM and DL plants decreased by 2 fold for Cr, 2.5 fold for As and ,1.3 and 3.6 for the
469 Cu respictivelly, whereas mineral mass of Cr and Cu in OMDL plants increased by 16% and 23% recpictivelly.
470 Differences between OM and OMDL plants were significant for Cr and Cu mineral masses. Lowest values of
471 As_{spw} and Cr_{spw} in OMDL soil were not reflected by Cr and As mineral mass of OMDL bean (Fig. 3) the
472 variance of Cu mineral masse was significant also between the contaminated soils (UNT, OM ,DL and
473 OM+DL) and the CTRL soil (p=0.00005).

474

475

476

477

478
479

Table 6: Dry weight (DW) of bean primary leaves BL (BLDW) and PTTE concentrations of bean primary leaves. Values are mean ± standard deviation (n=4).

traitments	Dry weight (DW) of bean primary leaves BL(BLDW) (g DWplant^{-1})	Foliar concentrations (mg. kg^{-1})			
		Cr	Cu	As	Zn
UNT	0.06±0.01	6.89 **a**±0.54	357 **a**±48.8	3.29 **a**±0.56	100 **a**±11.2
OM	0.11±0.00	1.58 **b**±0.03	142. **b**±23.7	0.65 **b**±0.01	52.2 **b**±4.43
DL	0.07±0.09	2.31 **b**±0.25	211 **b**±20.9	1.15 **b**±0.39	75.9 **b**±22.9
OMDL	0.10±0.01	5.37**ab**±0.30	260 **b**±15.8	1.55 a**b**±0.07	66.7 **b**±22.2
CTRL	0.13±0.00	1.18 **c**±0.14	16,0 **c**±2.80	0.19 **c**±0.00	44.7 **c**±6.34
Anova with CTRL	**NS**	**	**	**	*
Anova without CTRL	**NS**	**	**	**	**NS**

0.01<(*)P <0.05, 0.01>(**) p >0.001, (***) p< 0.0001, (NS) insignificant difference.

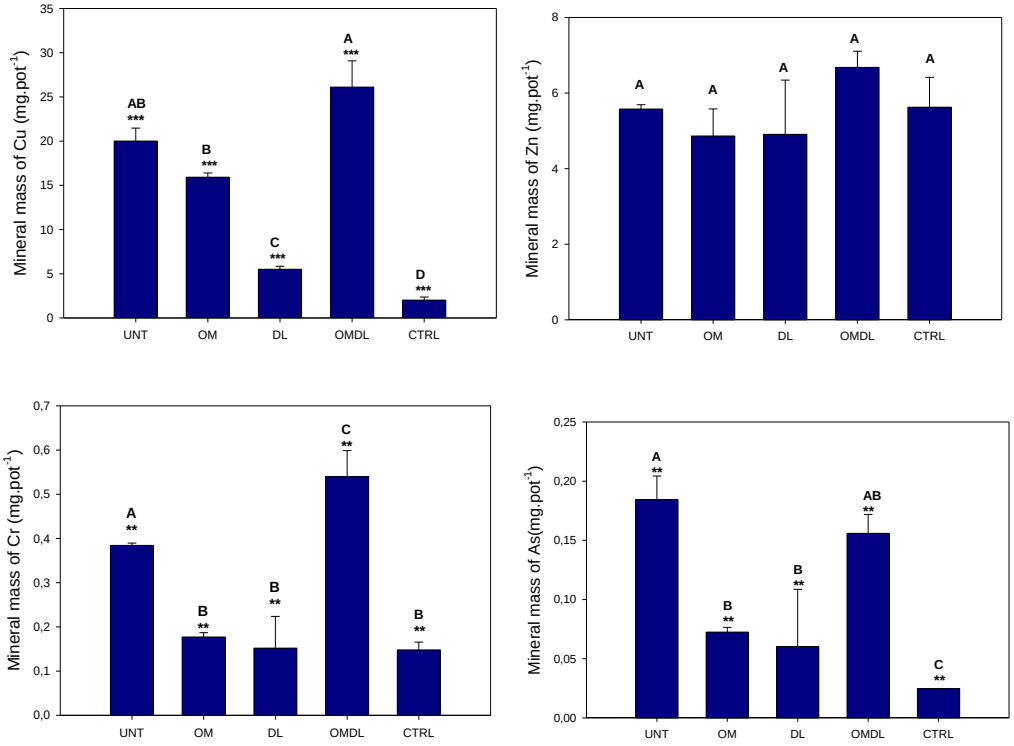

Figure 3: Mineral masses of Cu, Cr, Zn and As in the bean primary leaves. Values are mean ± standard deviation (n=4). Different letters on bar graphs indicate a significant difference (p<0.05)

4. Conclusion

The present work was focused on assessing the effectiveness of several aided-phytostabilisation options based on liming and the addition of compost to remediate PTTE in contaminated soils. Changes in mobility and phytoavailability of Cu, Zn, Cr and As were investigated at a wood preservation site in topsoils of field plots amended with DL and OM, singly and in combination, and phystabilized with mycorhizal trees, after four years. The OM incorporation into the contaminated soil decreased the Cu, Cr, Zn and As concentrations of bean primary leaves to the highest extent, but only their Cr and As mineral masses were reduced. The Cu

concentration in the SPWincreased for OM and OMDL soils whereas it slightly decreased for the DL soil. The Zn concentration in SPW significantly decreased only for the DL soil. The Cr concentration in SPW was enhanced for both DL and OM soils but slightly decreased for the OMDL soil. The As concentration in SPW increased for all amended soils, notably for the OM soil. The addition of OM and DL, singly and in combination, decreased the available fraction of Cu in soil determined by DGT. Mobilization of Cu from the soil to the SPW was slower than root uptake and soil amendments reduced the replenishment of Cu in SPW. Conversely, based on the DGT R ratio, Zn remobilization from the solid phases was enhanced in the DL soil.

In overall, data suggested the influence of both pH and DOM on metals and As concentrations in the SPW and their phytoavailability to dwarf beans. The DL addition promoted Cu and Zn sorption in relation with pH rise but enhanced remobilization of Cr and As, likely as chromates and arsenates from Fe oxides. Nevertheless foliar Cr and As concentrations in the DL beans were lower than in the UNT beans likely due to the decrease in soil Cu exposure, partial restoration of root growth and subsequent Cr and As storage in roots. Compost likely increased Cu–DOM complexes in the SPW, which are less available for root uptake, and consequently enhanced the DW yield of bean primary leaves. Subsequently, decreased soil Cu exposure in both OM-amended soils allowed roots to better control metals and As uptake and promote a dilution effect in the bean plant aerial parts.

In conlusion both aided-phytostabilisation options were found to be able to reduce the SPW concentrations or phytoavailability of PTTE and provide interesting technologies for the remeadiation of Cu and CCA salt contaminated soils.

Acknowledgements

Author thank Mrs Audrey Dufour (Cetrahe, University of Orléans, France) for the analysis of major cations and anions in the soil pore water and Dr Stanislas Strekopytov (Natural History Museum, London, UK) for the digestion of plants and subsequent mineral analysis. Dr M. Mench is grateful to ADEME, Department of Urban Browfields and Polluted Sites, Angers, France and the European Commission under the Seventh Framework Program for Research (FP7-KBBE-266124, GREENLAND) for financial support of the BIOGECO phytoremediation platform.

References

Alloway BJ (1995) Heavy metals in soils. Chapman & Hall, London.

Ashworth DJ, Alloway BJ (2007) Complexation of copper by sewage sludge-derived dissolved organic matter: Effects on soil sorption behaviour and plant uptake. Water Air and Soil Pollution 182 :187-196.

Baize D, Tercé M (2002) Les Éléments Traces Métalliques dans les Sols- Approches Fonctionnelles et Spatiales, INRA Éditions, Paris, France. 570pp.

Baize D (1997) Teneurs totales en éléments traces métalliques dans les sols (France). Références et stratégies d'interprétation. INRA Éditions, Paris, 410 p.

Balasoiu CF, Zagury GJ, Deschesnes L (2001) Partitioning and speciation of chromium, copper, and arsenic in CCA-contaminated soils: influence of soil composition. The Science of the Total Environment 280 (1–3): 239–255.

Banks M, Schwab A, Henderson C (2006) Leaching and reduction of chromium in soil as affected by soil organic content and plants. *Chemosphere* 62: 255-264.

Barnhart J (1997) Occurrences, uses, and properties of chromium. Regulatory Toxicology and Pharmacology 26 (1): S3–S7.

Basta NT, Gradwohl R, Snethen KL, Schroder JL (2001) Chemical immobilization of lead, zinc, and cadmium in smelter-contaminated soils using biosolids and rock phosphate. Journal of Environmental Quality 30 (4): 1222–1230.

Beesley L, Dickinson N (2011) Carbon and trace element fluxes in the pore water of an urban soil following greenwaste compost, woody and biochar amendments, inoculated with the earthworm Lumbricus terrestris. Soil Biology & Biochemistry 43: 188-196.

Beesley L, Dickinson NM (2010) Carbon and trace element mobility in an urban soil amended with green waste compost. Journal of Soils and Sediments 10: 215-222.

Beesley L, Moreno-Jiménez E, Gomez-Eyles JL (2010) Effects of biochar and greenwaste compost amendments on mobility, bioavailability and toxicity of inorganic and organic contaminants in a multi-element polluted soil. Environ Pollut 158: 2282–2287.

Bes C (2008) Phytoremédiation des sols d'un site de traitement du bois contaminé par le cuivre. Ph.D. Dissertation, Université de Bordeaux 1, Talence, France

Bes C, Mench M (2008) Remediation of copper-contaminated topsoils from a wood treatment facility using in situ stabilization. Environmental Pollution, 156: 1128-1138.

Bes CM, Mench M, Aulen M, Gaste H, Taberly J (2010) Spatial variation of plant communities and shoot Cu concentrations of plant species at a timber treatment site. Plant Soil 330: 267–80.

Bes CM, Jaunatre R, Mench M (2013) Seed bank of Cu-contaminated topsoils at a wood preservation site: impacts of copper and compost on seed germination. Environmental Monitoring and Assessment 185: 2039-2053

Bolan NS, Duraisamy VP (2003) Role of inorganic and organic soil amendments on immobilisation and phytoavailability of heavy metals: a review involving specific case studies. Aust. J. Soil Res. 41: 533–555.

Bolan NS, Adriano DC, Natesan R, Koo BJ (2003) Effects of organic amendments on the reduction and phytoavailability of chromate in mineral soil. Journal of Environmental Quality 32: 120-128.

Brallier S, Harrison RB, Henry CL, Dongsen X (1996) Liming effects on availability of Cd, Cu, Ni, and Zn in soil amended with sewage sludge 16 years previously. Water Air Soil Pollut. 86: 195-206.

Brown S, Chaney R, Hallfrisch J, Ryan JA, Berti WR (2004) In situ treatments to reduce the phyto and bioavailability of lead, zinc and cadmium. Journal of Environmental Quality 33: 522-531.

570 Brown S, Christensen B, Lombi E, McLaughlin M, McGrath S, Colpaert J, Vangronsveld J (2005) An inter-
571 laboratory study to test the ability of amendments to reduce the availability of Cd, Pb and Zn in situ.
572 Environ. Pollut. 138: 34–45.
573 Carey PL, McLaren RG, Adams JA (1996) Sorption of cupric, dichromate and arsenate ions in some New
574 Zealand soils, Water, Air, and Soil Pollut. 87: 189–203.
575 Cattani I, Fragoulis G, Boccelli R E, Capri E (2006) Copper bioavailability in the rhizosphere of maize (Zea
576 mays L.) grown in two Italian soils. Chemosphere 64: 1972–1979.
577 Chaignon V, Hinsinger P (2002) Fe-deficiency increases Cu acquisition by wheat cropped in a Cu-contaminated
578 vineyard soil. New Phytol. 154: 121–130.
579 Chaignon V, Hinsinger P (2003) A biotest for evaluating copper bioavailability to plants in a contaminated soil.
580 J. Environ. Qual. 32: 824–833.
581 Chen Z, Cai Y, Solo-Gabriele H, Snyder GH, Cisar JL (2006) Interactions of arsenic and the dissolved
582 substances derived from turf soils. *Environ Sci Technol*; 40: 4659–4665.
583 Chirenje T, Ma LQ (1999) Effects of acidification on trace metal mobility in a papermill ash amended soil. J.
584 Environ. Qual. 28 : 760–766.
585 Ciccu R, Ghiani M, Serci A, Fadda S, Peretti R, Zucca A (2003) Heavy metal immobilization in the mining-
586 contaminated soils using various industrial wastes. Minerals Engineering 16: 187 192.
587 Clemente R, Bernal MP (2006) Fractionation of heavy metals and distribution of organic carbon in two
588 contaminated soils amended with humic acids. Chemosphere 64: 1264-1273.
589 Davison W, Fones G, Harper M, Teasdale P, Zhang H (2000) Dialysis DET and DGT: in Situ Diffusional
590 Techniques for Studying Water, Sediments and Soils. In: Buffle J, Horvai G. (Eds.). In Situ Chemical
591 Analysis in Aquatic Systems, Wiley. 495-569.
592 Dobran S, Zagury GJ (2006) Arsenic speciation and mobilization in CCA-contaminated soils: Influence of
593 organic matter content. *Science of the Total Environment,* 364: 239-250.
594 Docekal B, Rezacova-Smetkova V, Docekalova H (2005) Effect of humic acid on metal uptake measured by
595 diffusive gradients in thin films technique. Chemical Papers-Chemicke Zvesti 59: 298 303.
596 Ernstberger H, Davison W, Zhang H, Tye A, Young S (2002) Measurement and dynamic modeling of trace
597 metal mobilization in soils using DGT and DIFS. Env. Sci. Technol. 36: 349-354.
598 Ernstberger H, Zhang H, DavisonW (2002) Determination of chromium speciation in natural systems using
599 DGT, *Anal. Bioanal. Chem.* 373: 873–879.
600 Filius A, Streck T, Richter J (1998) Cadmium sorption and desorption in limed topsoils as influenced by pH:
601 Isotherms and simulated leaching. *J. Environ. Qual.* 27: 12–18.
602 Forsberg LS, Kleja DB, Greger M, Ledin S (2009) Effects of sewage sludge on solution chemistry and plant
603 uptake of Cu in sulphide mine tailings at different weathering stages. Appl Geochem. 24: 475–482.
604 Garrido F, Illera V, Garcia-Gonzalez MT (2005) Effect of the addition of gypsum- and lime-rich industrial by-
605 products on Cd, Cu and Pb availability and leachability in metal-spiked acid soils. Appl. Geochem. 20:
606 397–408.

607 Geebelen W, Adriano DC, Van Der Lelie D, Mench M, Carleer R, Clijsters H, Vangronsveld J (2003) Selected
608 bioavailability assays to test the efficacy of amendment-induced immobilization of lead in soils. Plant and
609 Soil. 249. 12: 217-228.
610 González I, García F, Del Moral F, Simón M (2012) Effectiveness of amendments on the spread and
611 phytotoxicity of contaminants in metal–arsenic polluted soil. Journal of Hazardous Materials 205–206:
612 72–80.
613 Hartley W, Dickinson, Riby P, Elizabeth Leese E, Morton J, Lepp NW (2010) Arsenic mobility and speciation
614 in a contaminated urban soil are affected by different methods of green waste compost application.
615 Environmental Pollution 158: 3560-3570.
616 Hinsinger P (2001a) Bioavailability of trace elements as related to root introduced chemical changes in the
617 rhizosphere. In: Gobran GR, Wenzel WW, Lombi E. (Eds.). Trace Elements in the Rhizosphere. CRC
618 Press, Boca Raton, London, New York, Washington DC, 25–41.
619 Hinsinger P (2001b) Bioavailability of soil inorganic P in the rhizosphere as affected by root-induced chemical
620 changes: a review. Plant Soil 237: 173–195.
621 Hinsinger P (1998) How do plant roots acquire mineral nutrients. Chemical processes involved in the
622 rhizosphere. Adv. Agron. 24: 225–265.
623 Houben D, Pircar J, Sonnet P (2012) Heavy metal immobilization by cost-effective amendments in a
624 contaminated soil: effects on metal leaching and phytoavailability. J Geochem Explor.
625 doi:10.1016/j.gexplo.2011.10.004
626 Hsu JH, Lo SL (2000) Characterization of copper, manganese and zinc in swine manure composts. J. Environ.
627 Qual. 29, 447–453.
628 Jardine PM, Fendorf SE, Mayes MA, Larsen IL, Brooks SC, Bailey WB (1999) Fate and transport of hexavalent
629 chromium in undisturbed heterogeneous soil. Environmental Science and Technology 33: 2939-2944.
630 Jiang J, Xu R, Wang Y, Zhao A (2008) The mechanism of chromate sorption by three variable charge
631 soils.*Chemosphere* 71: 1469-1475.
632 Kabata-Pendias A, Pendias H (2000) Trace Elements in Soils and Plants, third ed. CRC Press Inc., Boca Raton,
633 USA.
634 Kimbrough DE, Cohen Y, Winer AM, Creelman L, Mabuni C (1999) A critical assessment of chromium in the
635 environment. Environmental Science and Technology 29, 1046.
636 Kumpiene J, Ore S, Renella G, Mench M, Lagerkvist A, Maurice C (2006) Assessment of zerovalent iron for
637 stabilization of chromium, copper, and arsenic in soil. Environmental Pollution 144: 62-69.
638 Kumpiene J, Lagerkvist A, Maurice C (2008) Stabilization of As, Cr, Cu, Pb and Zn in soil using amendments:
639 a review. Waste Management 28: 215–225.
640 Lagomarsino A, Mench M, Marabottini R, Pignataro A, Grego S, Renella G, Stazi S R (2011) Copper
641 distribution and hydrolase activities in a contaminated soil amended with dolomitic limestone and
642 compost. Ecotoxicology and Environmental Safety 74: 2013-2019.
643 Lee SH, Lee JS, Choi YJ, Kim JG (2009) In situ stabilization of cadmium- lead- and zinc-contaminated soil
644 using various amendments. Chemosphere, 77: 1069–1075

645 Lombi E, Sletten RS, Wenzel WW (2000) Sequentially extracted arsenic from different size fractions of
646 contaminated soils. *Water Air Soil Pollut*; 124: 319– 32.
647 Lombi E, Wenzel WW, Goran GR, Adriano DC (2001) Dependency of phytoavailability of metals on
648 indigenous and induced rhizosphere processes: a review. In: Gobran GR, Wenzel WW, Lombi E (Eds.).
649 Trace elements in the Rhizosphere. CRC Press, Boca Raton, London, New York, Washington DC, 4-23.
650 Lombi E, Zhao FJ, Wieshammer G, Zhang G, McGrath SP (2002) In situ fixation of metals in soils using
651 bauxite residue: biological effects. Environmental Pollution 118: 445- 452.
652 Macnicol RD, Beckett PHT (1980) Critical tissue concentrations of potentially toxic elements Plant Soil, 85:
653 107–129
654 McBride MB, Martinez CE (2000) Copper phytotoxicity in a contaminated soil: remediation tests with
655 adsorptive materials. Environmental Science & Technology 34: 4386–4391.
656 Mench M, Bert V, Schwitzguébel JP, Lepp n, Schröder P, Gawronski S, Vangronsveld J (2010) Successes and
657 limitations of phytotechnologies at field scale: Outcomes, assessment and outlook from COST Action
658 859. Journal Soils Sediments 10: 1039-1070
659 Mench M, Bes C (2009) Assessment of ecotoxicity of topsoils from a wood treatment site. Pedosphere. 19:143–
660 55.
661 Mench M, Bussiere S, Boisson J, Castaing E, Vangronsveld J, Ruttens A, De Koe T, Bleeker P, Assuncao A,
662 Manceau A (2003) Progress in remediation and revegetation of the barren Jales gold mine spoil after in
663 situ treatment. Plant and Soil 249: 187–202.
664 Morel JL, Echevarria G, Goncharova N (2006) Phytoremediation of Metal-Contaminated Soils. NATO Science
665 Series: IV: Earth and Environmental Sciences.
666 Palmer CD, Puls RW (1984) Natural Attenuation of Hexavalent Chromium in Groundwater and Soils. EPA
667 Ground Water Issue, EPA/540/5e94/505.
668 Pantsar-Kallio M, Reinikainen SP, Oksanen M (2001) Interactions of soil components and their effects on
669 speciation of chromium in soils. Analytica Chimica Acta 439 : 9–17.
670 Porter SK, Scheckel KG, Impellitteri CA, Ryan JA (2004) Toxic metals in the environment: thermodynamic
671 considerations for possible immobilisation strategies for Pb, Cd, As, and Hg. Critical Reviews in
672 *Environmental Science and Technology* 34: 495–604.
673 Rai UN, Pandey S, Sinha S, Singh A, Saxena R, Gupta DK (2004) Revegetating fly ash landfills with Prosopis
674 juliflora L.: impact of different amendments and Rhizobium inoculation. Environment International 30:
675 293–300.
676 Ross SM (1994) Toxic Metals in Soil Plant Systems. John Wiley & Sons, Chichester, UK.
677 Ruttens A, Colpaert J, Bradl M, Boisson J, Carleer R, Vangronsveld J (2006) Phytostabilization of a metal
678 contaminated sandy soil. II: Influence of compost and/or inorganic metal immobilizing soil amendments
679 on metal leaching. Environ. Pollut. 144 (2): 542–548.
680 Sadiq M (1997) Arsenic chemistry in soils: An overview of thermodynamic predictions and field observations,
681 *Water, Air, and Soil Pollut.* 93: 117–136.

682 Saha J.K, Mondal A.K, Hazra G.C, Mandal B (1991) Depth wise distribution of copper fractions in some ultisol.
683 Soil Sci. 151: 452–458.
684 Sanchez-Monedero MA, Roig A, Cegarra J, Bernal MP, Noguera P, Abad M, Anton A (2004) Composts as
685 media constituents for vegetable transplant production, Compost Sci. Util. 12: 161-168.
686 Sauve S, Mc Bride MB, Norvell WA and Hendershot WH (1997) Copper solubility and speciation of in situ
687 contaminated soils: effects of copper level, pH and organic matter. Water, Air and Soil Pollution. 100,
688 133-149.
689 Sauve S (2003) Modelling trace element exposure and effects on plants. In Risk assessment and sustainable land
690 management using plants in trace element-contaminated soils. Ed. M Mench, Mocquot, B.69-70, Centre
691 INRA Bordeaux-Aquitaine, Villenave d'Ornon, France.
692 Seaman JC, Hutchison JM, Jackson BP, Vulava VM (2003) In situ treatment of metals in contaminated soils
693 with phytate. Journal of Environmental Quality 32: 153–161.
694 Su D C, Wong J W C (2004) Phytoremediation potential of oilseed rape (*Brassica juncea*) for cadmium
695 contaminated soil. *Bulletin of Environmental Contamination and Toxicology*, 72(4): 991–998.
696 Tandy S, Schulin R, Nowack B (2006) Uptake of metals during chelant-assisted phytoextraction with EDDS
697 related to the solubilized metal concentration. Environ Sci Technol. 40: 2753–2758.
698 Thakali S, Allen HE, Di Toro DM, Ponizovsky AA, Rooney CP, Zhao FJ, McGrath SP (2006) A Terrestrial
699 Biotic Ligand Model. 1. Development and application to Cu and Ni toxicities to barley root elongation in
700 soils. Environmental Science & Technology 40: 7085-7093.
701 Zhang H, Zhao FJ, Sun B, Davison W, McGrath SP (2001) A new method to measure effective soil solution
702 concentration predicts copper availability to plants. Environ. Sci. Technol. 35: 2602 -260.
703